蝶と蛾 Trans. lepid. Soc. Japan 58 (3): 367-370, June 2007

# The germ-line chromosomes of *Ussuriana stygiana* (Butler) and *Coreana raphaelis* (Oberthür) (Lepidoptera, Lycaenidae) from Japan

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**Abstract** Spermatogonial and spermatocyte metaphase chromosomes of *Ussuriana stygiana* and *Coreana raphaelis* from Japan were examined. Chromosome number of the former species (U. sty-giana) was found to be 2n=102, n, 51, and of the latter (C. raphaelis) n, 51. Consequently, the haploid numbers of both species are the same.

Key words Chromosomes, Lycaenidae, Lepidoptera.

#### Introduction

The genera *Ussuriana* and *Coreana* belong to the common lower branch of the supposed phylogenetic tree of the so-called "*Zephyrus*", Theclini, Lycaenidae (Shirôzu and Saigusa, 1980), the former including three species, *U. michaelis* (including *gabrielis* sensu Fujioka (1992)), *U. fani* and *U. stygiana* (Mr S. Koiwaya's personal communication), and the latter only one, *C. raphaelis*.

On the chromosome numbers of the two genera, Abe (1997) reported those of U. michaelis as being n=41, and C. raphaelis n, 51, while Maeki and Remington (1960) and Robinson (1971) reported that of U. stygiana as being n=47.

From our present study, we find that the sample specimens of *U. stygiana* have 51 haploid chromosomes. This result disagrees with Maeki and Remington (1960) and Robinson (1971).

#### Materials and methods

Testes taken from laboratory-reared young pupae and mature larvae were used as material. The collecting localities of the sample specimens examined are given in the Table 1.

The testes of two species were squashed after staining with aceto-orcein.

Several testes of *U. stygiana* were prepared according to Crozier (1968).

The testes were fixed in Carnoy's solution (3: 1), carefully dissociated in aceto-acid, and then air-dried over the slide glass. The chromosomes were stained with Giemsa (6%).

The testes of one pupa and one larva of C. raphaelis from Asamushi, Aomori Pref. were fixed in P. F. A-3 mixture and sectioned (8  $\mu$ m) by the conventional method. The sections were stained by Heidenhain's iron-haematoxylin.

#### Observations and remarks

1. Ussuriana stygiana

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Table 1. Chromosome number of Ussuriana stygiana and Coreana raphaelis.

Species	Locality	Nos. and stages of males	Testis-technique used
Ussuriana stygiana	Asamushi, Aomori Pref.	1 pupa & 1 larva	Sectioning
	Minmaya, Aomori Pref.	2 pupae	Squashing
	Lake Juniko, Aomori Pref.	4 pupae	CROZIER
Coreana raphaelis	Kuji, Iwate Pref.	3 pupae	Sectioning
	Shinjo, Yamagata Pref.	2 pupae	Sectioning

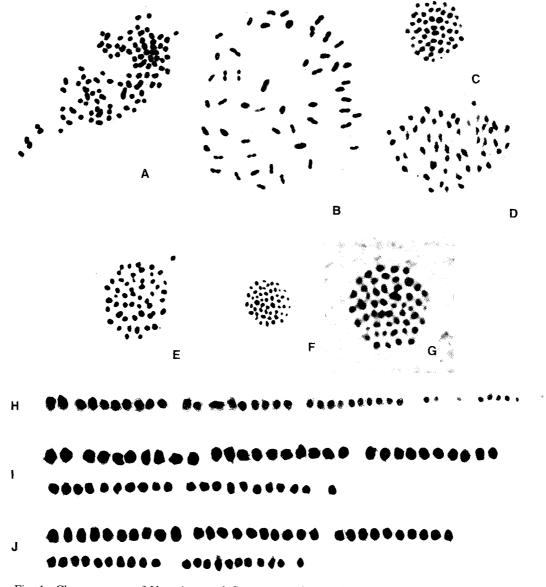


Fig. 1. Chromosomes of *Ussuriana* and *Coreana* species. A-D, I. *U. stygiana* (A: Spermatogonial metaphase, 2n, 102. Crozier. B: First-metaphase, n, 51. Crozier. C: Ditto, n, 51. Squash. D: Second-metaphase, n, 51. Crozier. I: n, 51.). E-G. C. raphaelis (E: First-metaphase, n, 51. Squash. F: Second-metaphase, n, 51. Squash. G: First-metaphase, n, 51. Section.). H. *U. michaelis*, n, 41. J. C. raphaelis, n, 51.

The sample specimens from three different localities, Asamushi, Minmaya, Aomori Pref. and Lake Juniko, Aomori Pref. show consistently 2n=102, n=51, with no variation observed in the haploid number. Determination of the diploid chromosome number is based on the counting of 26 spermatogonia (Crozier), and determination of the haploid chromosome number is based both on the counting of 88 primary spermatocytes (46 in sections, 24 in squashes, and 18 in Crozier) and on the counting of 32 secondary spermatocytes (21 in sections and 11 in squashes). The diploid chromosome of *Ussuriana stygiana* consists of four peculiar elements which are distinguishable from the others by their large size (Fig. 1A), and two elements in haploidity (Figs 1B, C, D).

### 2. Coreana raphaelis

Three pupae from Kuji, Iwate Pref. were examined: the testes of one of them were sectioned and those of the other two were squashed (Figs 1E, F). The testes of two pupae from Shinjo, Yamagata Pref. were sectioned in the same way (Fig. 1G).

The haploid chromosome number shows n=51 with no exception.

The number of cells observed is 118 in primary spermatocytes (79 in sections and 39 in squashes), and 41 in secondary spermatocytes (24 in sections and 17 squashes). One large element appears in haploidity.

#### **Discussion**

All the sample specimens of *Ussuriana stygiana* obtained from three localities consistently exhibit 2n=102, n=51, showing a difference from the report of n=47 for this species by Maeki and Remington (1960) and Robinson (1971). In order to clarify this discrepancy in the haploid number, the speciation history of *U. stygiana* should be taken into consideration. In the present study, however, because of the scantiness of data on geographic populations of this species and the consequent lack of cytological information, it is difficult to discuss this discrepancy fully.

In Fig. 2 are shown the tentative karyotypes of the haploid chromosomes of three species (*U. michaelis*, *U. stygiana*, *C. raphaelis*) in order of their size.

Although the two species, U. michaelis and U. stygiana, have different haploid chromosome numbers, n=41, 51, respectively, they are quite similar in the fact that they have two large chromosomes which are discernible from other chromosomes (Figs 1H, I). On the other hand, C. raphaelis, which has the same haploid chromosome n=51 as U. stygiana, has only one large chromosome (Fig. 1J). As far as the tentative karyotypes are concerned, U. stygiana is supposed to be more related to C. raphaelis than to U. michaelis.

#### Acknowledgements

We are indebted to Messrs K. Oda of Iwate Pref., Y. Kikuchi of Aomori Pref., and M. Kushibiki of Saitama Pref., for providing us with valuable research materials. We also express our cordial thanks to Mr S. Koiwaya, Miyazaki, for critical reading of classification of the genus *Ussuriana*.

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#### 摘 要

ウラキンシジミとチョウセンアカシジミの染色体(阿部 東・工藤貢次・斉藤和夫)

ウラキンシジミ Ussuriana stygiana とチョウセンアカシジミ Coreana raphaelis の染色体を調査した.

ウラキンシジミ 2n=102 (大型4を含む), n=51 (大型2を含む) チョウセンアカシジミ n=51 (大型1を含む)

である. ウラキンシジミについては Maeki & Remington (1960) による n=47 の報告があるが, 今回の結果はこの報告と異なっていた.

(Accepted November 11, 2004)

Published by the Lepidopterological Society of Japan, 5-20, Motoyokoyama 2, Hachioji, Tokyo, 192-0063 Japan